AMENDMENT TO THE CLAIMS

This listing of claims will replace all prior versions, and listings, of claims in the application:

Listing of Claims:

Claims 1-154 (canceled).

155. (previously presented) A purified polynucleotide comprising a nucleotide sequence encoding a polypeptide that comprises an amino acid sequence at least 95% identical to a fragment of the Asp2 protein amino acid sequence of Figure 3 (SEQ ID NO: 4),

wherein said fragment is a continuous fragment of the Asp2 protein that includes the aspartyl protease active site tripeptides DTG and DSG shown in Figure 3 and exhibits aspartyl protease activity involved in processing APP into amyloid beta,

wherein the polypeptide lacks a transmembrane domain, and

wherein the polypeptide exhibits aspartyl protease activity involved in processing APP into amyloid beta.

156. (previously presented) A purified polynucleotide comprising a nucleotide sequence that hybridizes under the following stringent hybridization conditions to the complement of SEQ ID NO: 3:

(1) hybridization at 42°C in a hybridization buffer comprising 6x SSC and 0.1% SDS, and

(2) washing at 65°C in a wash solution comprising 1x SSC and 0.1% SDS;

wherein said nucleotide sequence encodes a polypeptide that lacks a transmembrane domain and exhibits aspartyl protease activity involved in processing APP into amyloid beta.

Claims 157 - 158 (canceled)

159. (currently amended) A purified polynucleotide according to ~~any one of claims 155-158~~ claim 155 or 156, wherein said polynucleotide encodes a polypeptide that includes a heterologous peptide tag.

160. (currently amended) A purified polynucleotide according to ~~any one of claims 155-158~~ claim 155 or 156 that is a cDNA.

161. (currently amended) A vector comprising a polynucleotide of ~~any one of claims 155-158~~ claim 155 or 156.

162. (currently amended) A host cell transformed or transfected with a polynucleotide of ~~any one of claims 155-158~~ claim 155 or 156.

163. (previously presented) A host cell according to claim 162 that is a mammalian cell.

164. (previously presented) A host cell according to claim 163 derived from a human cell line.

165. (currently amended) An expression vector comprising a polynucleotide of ~~any one of claims 155-158~~ claim 155 or 156, wherein the polynucleotide is operably linked to a heterologous expression control sequence.

166. (previously presented) A host cell transformed or transfected with the vector of claim 165.

167. (previously presented) A host cell of claim 166 that is a mammalian cell.

168. (previously presented) A vector of claim 165 wherein the polynucleotide is operably linked to a heterologous control sequence for expression in a mammalian host cell.

169. (previously presented) A vector of claim 165, wherein the polynucleotide is operably linked to a heterologous control sequence for expression in an insect host cell.

170. (withdrawn) A method for identifying agents that inhibit the aspartyl protease activity of the polypeptide encoded by a polynucleotide of claim ~~151~~ 155 or 156, comprising the steps of:

(a) contacting amyloid precursor protein (APP) and the polypeptide in the presence and absence of a test agent;

(b) measuring the aspartyl protease activity of the polypeptide in the presence and absence of the test agent; and

(c) comparing the aspartyl protease activity of the polypeptide in the presence of the test agent to the activity in the absence of the test agent to identify an agent that inhibits the aspartyl protease activity of Hu-Asp2, wherein reduced aspartyl protease activity in the presence of the test agent identifies an agent that inhibits aspartyl protease activity of the polypeptide.

171. (withdrawn) A method according to claim 170 wherein the polypeptide is encoded by a nucleotide sequence selected from the group consisting of:

(a) a nucleotide sequence encoding the Hu-Asp2 amino acid sequence set forth in Figure 3 (SEQ ID NO: 4);

(b) a nucleotide sequence encoding a continuous fragment of the amino acid sequence of Figure 3(SEQ ID NO: 4), wherein said fragment includes the aspartyl protease active site tripeptides DTG and DSG shown in Figure 3 and exhibits aspartyl protease activity involved in processing APP into amyloid beta; and

(c) a nucleotide sequence that hybridizes under the following stringent hybridization conditions to the complement of a of SEQ ID NO: 3:

(1) hybridization at 42°C in a hybridization buffer comprising 6x SSC and 0.1% SDS, and

(2) washing at 65°C in a wash solution comprising 1x SSC and 0.1% SDS.

172. (withdrawn) A method according to claim 170 wherein the polypeptide comprises the amino acid sequence set forth in SEQ ID NO: 4.

173. (withdrawn) A method according to claim 170, wherein the polypeptide comprises a continuous fragment of the amino acid sequence of Figure 3 (SEQ ID NO: 4) wherein said fragment includes the aspartyl protease active site tripeptides DTG and DSG shown in Figure 3 and exhibits aspartyl protease activity involved in processing APP into amyloid beta.

174. (withdrawn) A method according to claim 170, wherein the APP comprises a carboxy-terminal di-lysine (KK) and wherein the contacting comprises growing a host cell that expresses the APP in the presence and absence of the test agent.

175. (withdrawn) A method according to claim 174, wherein the host cell has been transformed or transfected with a polynucleotide comprising a nucleotide sequence that encodes a Hu-Asp2 polypeptide, wherein said nucleotide sequence is selected from the group consisting of

(a) a nucleotide sequence encoding the amino acid sequence set forth in Figure 3 (SEQ ID NO: 4);

(b) a nucleotide sequence encoding a continuous fragment of the amino acid sequence set forth in Figure 3 (SEQ ID NO: 4), wherein said fragment includes the aspartyl protease active site tripeptides DTG and DSG shown in Figure 3 and exhibits aspartyl protease activity involved in processing APP into amyloid beta; and

(c) a nucleotide sequence of a polynucleotide that hybridizes under the following stringent hybridization conditions to the complement of SEQ ID NO: 3:

(1) hybridization at 42°C in a hybridization buffer comprising 6x SSC and 0.1% SDS, and

(2) washing at 65°C in a wash solution comprising 1x SSC and 0.1% SDS.

176. (withdrawn) A method for identifying agents that inhibit the APP processing activity of human Asp2 aspartyl protease, comprising the steps of:

(a) contacting a polypeptide comprising the amino acid sequence set forth in Figure 3 (SEQ ID NO: 4) or a continuous fragment thereof and amyloid precursor protein (APP) in the presence and absence of a test agent, wherein the contacting comprises growing a host cell transformed or transfected with a polynucleotide comprising a nucleotide sequence encoding the Asp2 polypeptide in the presence and absence of the test agent,

wherein said fragment is a continuous fragment of the Asp2 protein that includes the aspartyl protease active site tripeptides DTG and DSG shown in Figure 3 and exhibits aspartyl protease activity involved in processing APP into amyloid beta,

(b) measuring the APP processing activity of the polypeptide in the presence and absence of the test agent; and

(c) comparing the APP processing activity of the polypeptide in the presence of the test agent to the activity in the absence of the test agent to identify an agent that inhibits the APP processing activity of the polypeptide, wherein reduced activity in the presence of the test agent identifies an agent that inhibits the APP processing activity of the polypeptide.

177. (withdrawn) A method according to claim 176, wherein the host cell expresses APP.

178. (withdrawn) A method according to claim 177, wherein the host cell expresses an APP having an amino acid sequence that includes a carboxy-terminal di-lysine.

179. (withdrawn) A method according to any one of claims 176-178, wherein the host cell is a human embryonic kidney cell line 293 (HEK293) cell.

180. (withdrawn) A method according to claim 176, wherein the host cell comprises a vector that comprises the polynucleotide.

181. (withdrawn) A method according to any one of claims 170, 171, 174, 175 or 176 wherein the APP comprises the Swedish mutation (K→N, M→L) adjacent to the β-secretase processing site.

182. (withdrawn) A method according to any one of claims 174, 175 or 176 wherein the measuring step comprises measuring the production of amyloid beta peptide by the host cell in the presence and absence of the test agent.

183. (withdrawn) A method for identifying an agent that inhibits APP processing activity of the polypeptide <u>encoded by a polynucleotide</u> of ~~any one of claims 151-153~~ <u>claim 155 or 156</u>, comprising steps of:

(a) contacting the polypeptide with an APP substrate, in the presence and absence of a test agent;

(b) measuring the proteolytic processing of the APP substrate by the polypeptide in the presence and absence of the test agent; and

(c) comparing the proteolytic processing of the APP substrate by the polypeptide in the presence and absence of the test agent to identify an agent that inhibits the APP

processing activity of the polypeptide, wherein reduced proteolytic processing of the APP substrate by the polypeptide in the presence of the test agent identifies an agent that inhibits APP processing activity of the polypeptide.

184. (withdrawn) A method according to claim 183, wherein the APP substrate is a peptide comprising a β-secretase cleavage site of APP.

185. (withdrawn) A method according to claim 184, wherein the β-secretase cleavage site comprises the formula P2-P1-P1'-P2', wherein

P2 is an amino acid selected from K and N;

P1 is an amino acid selected from M and L;

P1' is the amino acid D; and

P2' is the amino acid A.

186. (withdrawn) A method according to claim 185, wherein the peptide comprises the amino acid sequence KMDA (SEQ ID NO: 64, positions 4-7).

187. (withdrawn) A method according to claim 185, wherein the peptide comprises the amino acid sequence EVKMDAEF (SEQ ID NO: 67).

188. (withdrawn) A method according to claim 185, wherein the peptide comprises the amino acid sequence NLDA (SEQ ID NO: 66).

189. (withdrawn) An isolated nucleic acid comprising nucleotides encoding a biologically active human aspartyl protease containing a valine at a position which corresponds to position 130 of SEQ ID NO:4, or a conservative substitute therefor, which isolated nucleic acid hybridizes ~~under stringent wash conditions~~ to the complement of a nucleic acid encoding the amino acid sequence set forth in SEQ ID NO: 4 under the following stringent wash conditions:

(1) hybridization at 42°C in a hybridization buffer comprising 6x SSC and 0.1% SDS, and

(2) washing at 65°C in a wash solution comprising 1x SSC and 0.1% SDS.

190. (withdrawn) An isolated nucleic acid comprising nucleotides encoding a biologically active human aspartyl protease containing a valine at a position which corresponds to position 130 of SEQ ID NO:4, the sequence of which isolated nucleic acid is identical across its length to a sequence set forth in SEQ ID NO: 3.

191. (withdrawn) An isolated nucleic acid comprising nucleotides encoding a biologically active human aspartyl protease containing a valine at a position which corresponds to position 130 of SEQ ID NO:4, the sequence of which isolated nucleic acid is identical across its length to the sequence set forth in SEQ ID NO: 3.

192. (withdrawn) An isolated nucleic acid comprising nucleotides encoding a biologically active human aspartyl protease, the sequence of which isolated nucleic acid is identical to a sequence set forth within SEQ ID NO: 3.

193. (withdrawn) A vector comprising the isolated nucleic acid of any one of claims 189-192.

194. (withdrawn) A host cell comprising the vector of claim 193.

195. (withdrawn) An isolated biologically active human aspartyl protease containing a valine at a position which corresponds to position 130 of SEQ ID NO:4, which human aspartyl protease is encoded by a nucleic acid which hybridizes ~~under stringent wash conditions~~ to the complement of a nucleic acid encoding the amino acid sequence set forth in SEQ ID NO: 4 under the following stringent wash conditions:

(1) hybridization at 42°C in a hybridization buffer comprising 6x SSC and 0.1% SDS, and

(2) washing at 65°C in a wash solution comprising 1x SSC and 0.1% SDS.

196. (withdrawn) An isolated biologically active human aspartyl protease containing a valine at a position which corresponds to position 130 of SEQ ID NO: 4, which human aspartyl protease is encoded by a nucleic acid which is identical across its length to a sequence set forth in SEQ ID NO: 3.

197. (withdrawn) An isolated biologically active human aspartyl protease containing a valine at a position which corresponds to position 130 of SEQ ID NO: 4, which human aspartyl protease is encoded by a nucleic acid which is identical across its length to the sequence set forth in SEQ ID NO: 3.

198. (withdrawn) An isolated biologically active human aspartyl protease containing a valine at a position which corresponds to position 130 of SEQ ID NO: 4, which human aspartyl protease is encoded by a nucleic acid which is identical to a sequence set forth within SEQ ID NO: 3.

199. (withdrawn) An isolated polypeptide with aspartyl protease activity comprising an amino acid sequence which is identical across its length to a sequence in SEQ ID NO: 4.

200. (withdrawn) An isolated polynucleotide comprising a nucleotide sequence encoding a polypeptide with aspartyl protease activity, wherein said polypeptide comprises an amino acid sequence which is identical across its length to a sequence in SEQ ID NO: 4.